# Seasonal differences in the quality of shortfin glass eel, *Anguilla australis*, and subsequent effects on growth and survival in captivity

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**Abstract**

Where glass eel harvest from the wild is strictly controlled it is advantageous for aquaculturalists to ensure that the eels are of the highest quality. The quality of shortfin glass eels, *Anguilla australis* (Richardson), were compared among three sampling events over a migratory season (August, October and December 2005) for a river in northern New Zealand. Glass eel quality was assessed by measuring length, dry mass and lipid content, as well as subsequent growth and survival when reared under laboratory culture conditions in brackish water (17.5 ‰) maintained at 25° C over 70 days. The mean length, mass, lipid and survival of shortfin glass eels declined with the progression of the migratory season. There were positive relationships between all of the measures of quality for glass eels, i.e., mass and lipid content, mass and length, and length and lipid content. Mean specific growth rate (SGR) of shortfin glass eels captured at the start of the migratory season, August, and then cultured was significantly higher than for the subsequent two samples in October and December. These results indicate that aquaculturalists of this species in New Zealand should target the harvest of early season glass eels to produce the most effective subsequent aquaculture performance.

Key words: glass eel quality - survival - growth rate - aquaculture - *Anguilla australis.*

## Introduction

Studies in larval fish ecology have measured larval nutritional condition with the ultimate goal of assessing the potential for future growth and survival (Ferron & Leggett 1994; Elliott & Leggett 1998). Larvae in poor condition have been shown to be more affected by predation, disease, unfavorable environmental conditions and are less efficient at catching prey due to impaired swimming capability (Gagliano *et al.* 2007). In some fish, differences in size at

settlement have been shown to persist into early juvenile stages. In a laboratory study of winter flounder *Pseudopleuronectes americanus*, Chambers *et al.* (1988) found that size advantages gained from the larval stage were perpetuated in juveniles. Therefore, the size and nutritional condition of a fish can have important consequences for subsequent growth, survival, and successful transition to the next life history stage (Canino *et al.* 1991; Håkanson 1993).

Anguillid eels are catadromous, with adult eels spawning at sea followed by an extended marine larval phase that can last for more than a year in some species in temperate waters (Tesch 1977). The marine larval phase ends when glass eels arrive at the coast and begin to move into fresh waterways. Glass eels are a valuable resource that is harvested for restocking of waters where recruitment is limited, or as seed stock for eel aquaculture (Moriarty & Dekker 1997).

The European (*Anguilla anguilla*), Japanese (*A. japonica*) and American eels (*A. rostrata*) are extensively harvested during the glass eel migratory phase and there has been widespread concern about the diminishing recruitment of these species (Moriarty & Dekker 1997) resulting in renewed interest in the viability of aquaculture of other southern hemisphere species. Initial attempts to farm the shortfin eel and longfin eel (*A. dieffenbachii*, Gray) in New Zealand were not encouraging (McDowall 1995), but since then there have been major changes in farming technology, and the shortfin eel is now being farmed both extensively and intensively in Australia (Gooley *et al.* 1999).

The arrival times of the two main New Zealand species, the shortfin and longfin eel are generally well documented (Jellyman 1977, Jellyman 1979, Jellyman *et al.* 1999) and their arrival in waterways typically peak between September and October. Research on New Zealand and northern hemisphere Angullid species suggests there are marked differences in some of the qualities of glass eels which are associated with their time of arrival. Chisnall *et al.* (2002) researched the spatial and temporal variability in the length of glass eels in New Zealand, concluding that mean length of shortfin and longfin glass eels generally declined throughout the season, an effect documented for other temperate Anguillid species, i.e., the Australian shortfin (*A. australis*) and Australian longfin eel (*A. reinhardtii*) (Sloane 1984), European eel (Desaunay & Guerault 1997), American eel (Jessop 1998) and Japanese eel (Kawakami *et al.* 1999). Jellyman (1979) found a seasonal decline in the length, mass and condition (K) of shortfin and longfin migrating glass eels. For the Australian shortfin and longfin eel, the stage of pigmentation advanced as the season progressed, and length, weight and condition factor declined with advancing pigmentation (Sloane 1984). The nutritional status of the Japanese glass eel was analyzed according to RNA/DNA ratios, and these ratios significantly decreased in glass eels collected during the later months indicating that glass eels collected in the earlier months were in a better physiological condition (Kawakami *et al.* 1999).

While the seasonal trends in length, mass and body condition have been described for a number of Anguillid species, detailed information about the relationships between seasonal variations in lipid content is lacking, even though the importance of lipid utilization during the early life stages of fish is well documented and lipid availability has been associated with growth and survival in the post-settlement stages of a number of fish species (Rainuzzo *et al.* 1992). Lipid content is a commonly used indicator of larval fish condition; it is an important source of energy in developing fishes because of its very high energy to weight ratio compared to other forms of biochemical energy storage (McCormick 1998).



Since glass eels are collected for culture, understanding how glass eel quality (i.e., size, length, condition and lipid content) changes within a migratory season is a key research issue in fish culture and is likely to be of economic value to culturists where size and condition advantages secured in glass eels may translate into subsequent improvements in survival, growth rates and business profits (Ingram *et al.* 2001). There has been no research investigating the differences in the lipid content of glass eels across the migratory season, or whether differences in size or lipid content relate to subsequent performance in aquaculture.

The aim of this study was to determine the seasonal variability in size (length and dry mass) and lipid content for shortfin glass eel captured from the same river (Tairua River) at different times during a seasonal migration (August, October and December 2005). An additional aim was to ascertain if there is a link between the quality of glass eels (length, dry mass and lipid content) and their subsequent growth and survival under aquaculture conditions. These results should guide recommendations for aquaculturists on the timing of glass eel harvest and the potential for size sorting of harvested glass eels to make the best use of limited access to wild glass eel resource for aquaculture.

## Methods

### *Field Sampling*

Shortfin glass eels were collected by electric fishing in the lower reaches of the Tairua River (37°04'21.86"S, 175°46'08.88"E) on the east cost of the North Island of New Zealand on 22 August, 7 October and 1 December 2005. Catch rates varied across the season with 450 glass eels captured in August, 300 in October and 196 in December.

The glass eels were transported in thermally insulated plastic bins, containing 20 l of 13 °C freshwater, to the NIWA Bream Bay Aquaculture Park, north-eastern New Zealand. On arrival, glass eels were anaesthetised with AQUI-S® and categorized on the basis of pigmentation (Strubberg 1913); more lightly pigmented eels, stages 5B–6A23, were classified as newly arrived while those with more advanced pigmentation, stages >6A23, were classified as previously arrived. Only newly arrived glass eels (5B-6A23) were used in the experiment.

Glass eels were counted and 30 eels from each sample month were taken at random for condition analyses. The remaining eels were transferred into an 80 l holding tank and acclimated to 25 °C saltwater (35 ‰) over a 24 hour period (c. 0.5 °C and 1.5 ‰ per hour). A six day quarantine period followed as a precaution to assess survival, infectious disease and to help minimize the effects of transport stress. Saltwater was used during the quarantine period as it eliminates fresh water borne disease. Mortalities were counted daily. The eels were fed to satiation twice per day with frozen *Artemia*.

### *Condition Analyses*

Total length was measured from the most posterior point on the tail to the most anterior point on the head. Dry mass was recorded after freeze drying individual glass eels for 24 hours. Ten glass eels from each sample month (August, October and December) were analysed for total body lipid content (%) according to the method

of Bligh & Dyer (1959). Whole glass eels selected for lipid analysis were ground to a fine powder with a cryocrusher. Tissue samples were mixed thoroughly in separation funnels containing 5:10:4 ml chloroform: methanol: distilled water and left to stand for 6 hours, after which chloroform (10 ml) and distilled water salt solution (10 ml; 5 ‰) was added, vigorously shaken and left to stand overnight. The bottom chloroform layer was then drained into a round bottom flask and the solvent evaporated using a Rotoevaporator. Chloroform (1 ml) was added to the round bottom flask and contents drawn into pre weighed glass vials. The chloroform was evaporated off under pure nitrogen gas and the resulting residue weighed to allow the calculation of percentage lipid content of the glass eel.

*Growth and Survival*

After the six day quarantine period the glass eels were randomly divided into three groups and stocked into triplicate tanks at 140, 90 and 55 fish per tank for August, October and December respectively. Glass eels were reared in a flow through aquaculture system under identical experimental conditions. The eels were ongrown in 50 l black circular plastic tanks subjected to natural photoperiod. Each tank contained 30 l of filtered (7 µm) brackish water (17.5 ‰) maintained at 25 ± 3.5 °C with a water flow of approximately 3 l $min^{-1}$. Water temperature was checked daily with a thermometer and salinity was measured with a refractometer three times per week. These culture conditions were selected because this experiment was part of a larger project investigating the feasibility of culturing shortfin eels in brackish water.

The total biomass of fish in each tank was weighed weekly to calculate feeding rate. Fish were fed frozen *Artemia* enriched with fish oils from days 1 – 7 at a feeding rate calculated at 10 % b.w. (body-weight) $day^{-1}$. From days 8 - 14 frozen *Artemia* was reduced to 5 % b.w. $day^{-1}$ and a commercial weaning crumble diet (Proton) was introduced and fed to satiation. After day 14, frozen *Artemia* was removed from the diet and Proton was fed to eels at 10 % b.w. $day^{-1}$. Fish were hand fed three times per day at 0900, 1300 and 1700 hours. Mortalities were removed from the tanks daily throughout the course of the experiment. At the end of the experiment mean survival for each tank was calculated. Mean specific growth rate (SGR) was calculated as (ln final mean fish wt-ln initial mean fish wt.) / time (70 days) x 100 (Gooley *et al.* 1999).

*Statistical analysis*

The sampling and experimentation utilised balanced designs to allow for ANOVA methods (Sokal & Rohlf 2001). Differences in glass eel length, dry mass, lipid content, SGR and survival rate were analysed using a single factor analysis of variance. Significant differences between individual pairs of means were identified with Tukey's honestly significant difference method (Sokal & Rohlf 2001). Pearson's correlations were used to determine if there was a relationship between the lipid content of glass eels and their length or dry mass. Data were first checked for homogeneity of variance using the *F*-test. For all statistical tests significance was set at $P < 0.05$ and all mean values are presented as mean ± SE.

## Results

In total, 90 shortfin glass eels were measured for length and dry mass. The



**Table 1.** Mean (±SE) length, dry mass and lipid content of shortfin glass eels captured from the Tairua River in August, October and December of 2005. Mean values within the same row not sharing the same superscript are significantly different at $p < 0.05$ using Tukey's honestly significant difference test. ($n$=30 length & dry mass, $n$ = 10 lipid)

| Parameter | August | October | December |
|---|---|---|---|
| Length (mm) | $61.53 \pm 0.45^{a}$ | $58.50 \pm 0.36^{b}$ | $56.27 \pm 0.48^{c}$ |
| Dry Mass (mg) | $183.85 \pm 4.63^{a}$ | $160.92 \pm 3.65^{b}$ | $133.88 \pm 4.03^{c}$ |
| Lipid (%) | $14.41 \pm 0.59^{a}$ | $10.74 \pm 1.04^{b}$ | $11.69 \pm 0.93^{b}$ |

longest (65 mm) and heaviest (228 mg) glass eel was caught in August, and the shortest (50 mm) and lightest (99 mg) in December. Over all of the eels sampled the length ranged from 50 - 65 mm with a mean of 58.74 ± 0.34 mm; dry mass ranged from 99 - 228 mg with a mean of 158.77 ± 3.17 mg. There were significant differences in the mean length ($F_{2, 87}$ = 36.7, $p$ <0.0001) and dry mass ($F_{2, 87}$ = 38.7, $p$ <0.0001) of shortfin glass eels among months of the migratory season. The mean length and dry mass of shortfin glass eels declined progressively as the migratory season advanced from August to December (Table 1).

The lipid content of the 30 glass eels that were sampled over the migratory ranged from 2.38 – 22.07 % with a mean of 12.28 ± 0.57 %. The greatest lipid content came from a glass eel caught in October (22.07 %) and the lowest (2.38 %) in December. Mean lipid content differed among months ($F_{2, 27}$ = 4.73, $p$ < 0.05). The mean lipid content of shortfin glass eels was significantly higher in August than for glass eels sampled in either October or December (Table 1).

To establish if the three potential measures of glass eel quality were related they were compared with one another (i.e., eel dry mass × lipid content, length × dry mass, length × lipid content) using a Pearson's correlation test The tests revealed significant correlations between glass eel dry mass × lipid content (Figure 1A; $r$ = 0.88, $r^2$ = 0.78, df = 28, $p$ < 0.0001), length × dry mass (Figure 1B; $r$ = 0.65, $r^2$ = 0.42, df = 88, $p$ < 0.0001), and length × lipid content (Figure 1C; $r$ = 0.54, $r^2$ = 0.29, df = 28, $p$ < 0.0001).

The mean SGR of shortfin glass eels differed between sampling events ($F_{2, 6}$ = 63.82, $p$ <0.0001). The mean SGR of shortfin glass eels sampled in August (1.77 ± 0.07 % b.w. $day^{-1}$) was significantly higher than those sampled during October (1.21 ± 0.01 % b.w. $day^{-1}$) and December (1.20 ± 0.00 % b.w. $day^{-1}$) (Figure 2A). Glass eels captured in August on average grew roughly 50 % faster than in October and December.

The mean survival of shortfin glass eels differed significantly between sampling events ($F_{2, 6}$ = 203.68, $P$ <0.00001). The mean survival of shortfin glass eels declined as the migratory season progressed (August 83.78 ± 1.98 %, October 71.88 ± 0.90 % and December 42.93 ± 1.34 %) (Figure 2B). The survival rate of glass eels captured in August was approximately twice that of December. No measurements of dead eels were made, however, observations indicated that mortalities were always associated with the smallest glass eels with emaciated stomachs.

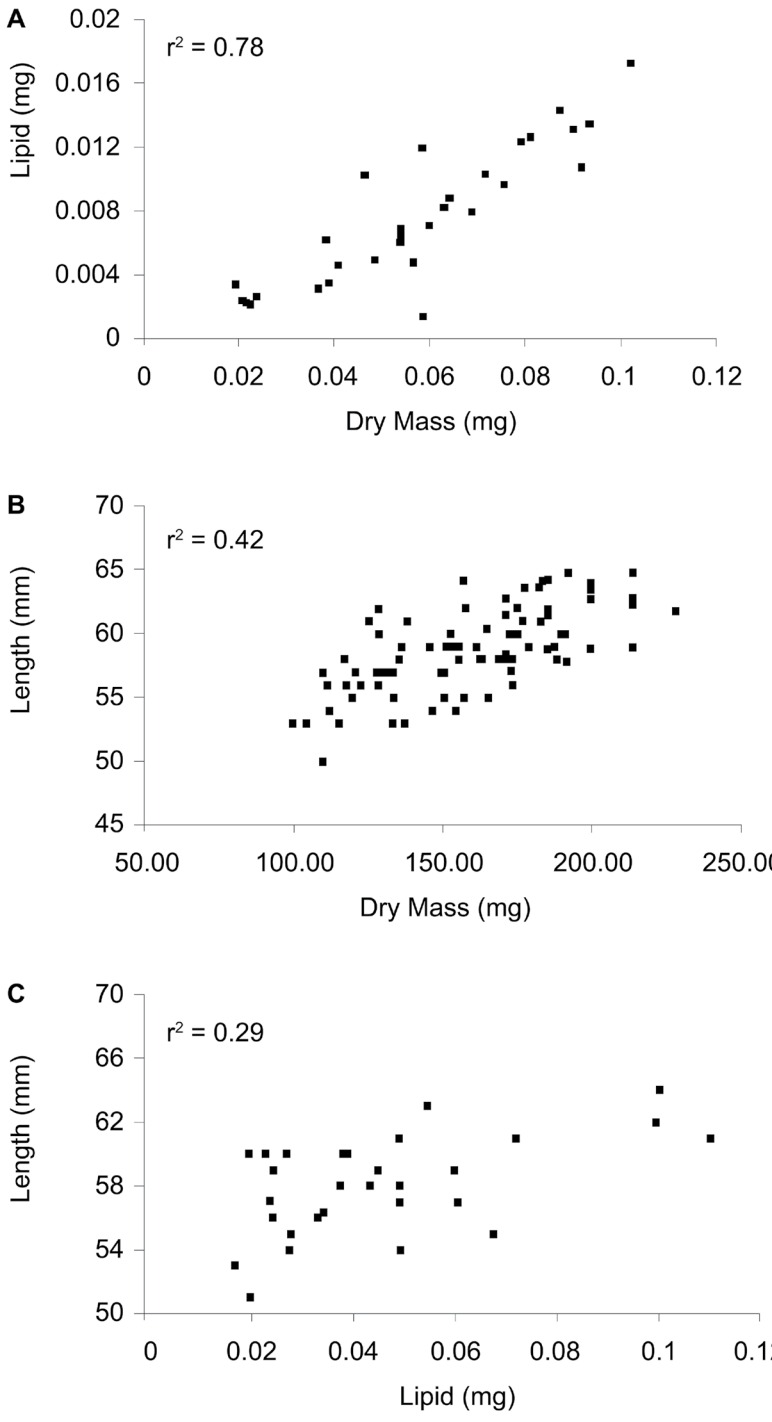


**Figure 1.** Scatter-plot showing the relationship between lipid content and dry mass (A), length and dry mass (B) and length and lipid content (C) for shortfin glass eels captured from the Tairua River during the 2005 freshwater migration. ($n$=30).



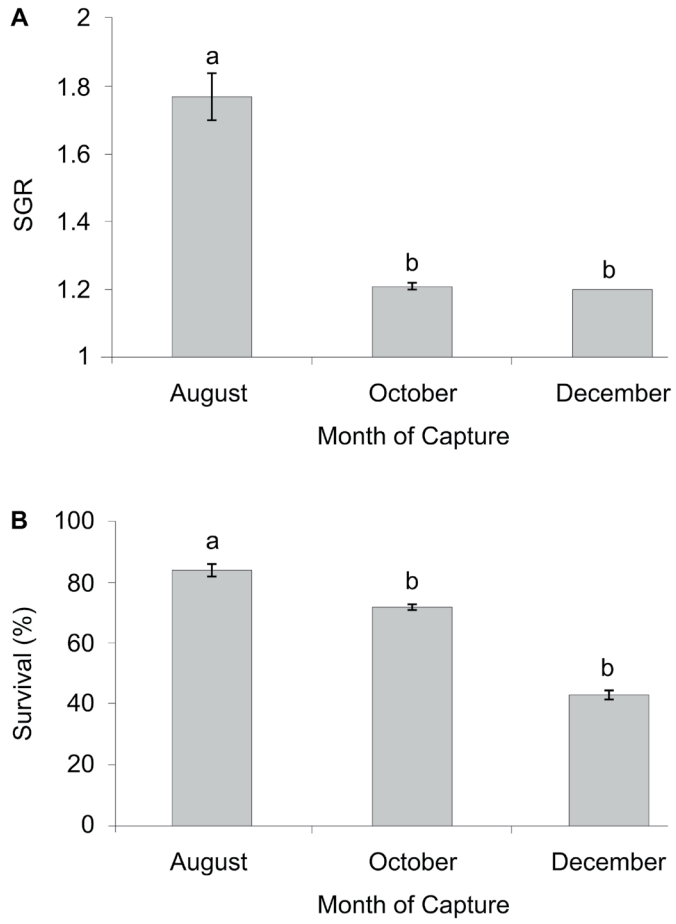


**Figure 2.** Mean (± SE) SGR (A) and survival rate (B) of shortfin glass eels reared at 25 ºC in 17.5 ‰ water for 70 days. Mean values not sharing the same superscript are significantly different at $P < 0.05$ using Tukey's honestly significant difference test ($n$ = 3).

## Discussion

Shortfin glass eels sampled at our study location declined in all measures of quality (length, dry mass, lipid content) from the initial sample early in the migratory season (August). This pattern was reflected in marked differences in the subsequent performance of the glass eels in experimental culture, with eels sampled early in the migratory season exhibiting higher growth and survival in culture compared to glass eels captured later in the season (October and December). This seasonal decline in glass eel performance was despite decreases in the experimental culture densities due to difficulties catching sufficient experimental eels as the season progressed. It could be expected that these decreased densities would improve survival in culture, the reverse of the experimental results (Degani, 1983, 1988; Degani *et al.* 1985; Roncarati *et al.* 1997).

Declines in glass eel size (length and wet weight) over the migratory season have been observed with other temperate Angullid species, including the Australian shortfin and the Australian longfin eel (Sloane 1984), the American eel (Jessop 1998) and the Japanese eel (Kawakami *et al.* 1999). However, this is the first time marked declines in the lipid content and dry mass of glass eel have been described

across the migratory season. A combination of variables has previously been used to explain the changes in the condition of glass eels, including variability on planktonic food supply for eel larvae and fluctuations in water temperature (Desaunay & Guerault 1997). Substantial levels of variability have also been found in the nutritional condition of reef fish at settlement, quantified by the total lipid, protein and carbohydrate composition of individual fish (McCormick & Molony 1993; Kerrigan 1996). Kerrigan (1996) found that for two species of tropical damsel fish, nutritional composition varied significantly both among lunar pulses of recruits and between years. Trends in lipid and protein composition between years and among pulses were similar for two species, suggesting that they were influenced by similar processes in the pelagic environment.

Two studies that have examined the interrelationship among various measures of condition of reef fish at settlement have found correlations among measures to be weak (McCormick & Molony 1993; Kerrigan 1996). In particular, fish length was weakly correlated to concentrations of energy storage, suggesting that no single measure of condition comprehensively describes a fish's potential to survive or compete. A number of other studies in temperate waters have reached a similar conclusion (Theilacker 1978; Neilson *et al.* 1986). In contrast, results from the present study indicate that all three potential measures of glass eel condition (i.e., length, dry mass and lipid content) show positive correlations with one another. These findings indicate that length and dry mass are reliable indicators of condition and that aquaculturalists should selectively harvest larger glass eels which would ultimately return higher SGR and survival rates.

It has also been suggested that the larger size and corresponding nutritional reserves of early season glass eels may help to compensate for the higher probability of seasonally adverse environmental conditions (lower water temperatures, high stream discharge, and low prey availability) early in the freshwater migration (Martin 1995; Jessop 1997, 1998). However, our results indicate poor growth and survival of the poorer quality late season glass eels when reared in experimental culture conditions with warm water and an abundance of high quality of food, which implies glass eels arriving later in the season have reduced capacity for subsequent survival. These late season glass eels did not have the energy reserves to transition effectively into the culturing environment and wean onto the artificial feed. The endogenous energy expenditure or stress due to transitioning from 11° C freshwater to the 25° C brackish water (17.5 ‰) may be sufficiently large to prevent survival of glass eels in poorer condition. The body mass of glass eels is known to decline as a result of metabolic use of energy reserves prior to initiation of post-larval feeding (Tesch 1977; Dutil *et al.* 1989), and Kawakami *et al.* (1999) suggests reductions in the weight of Japanese glass eels correspond to a decline in endogenous nutrition status, especially stored lipid. Therefore, it is likely that eels with larger energy reserves will be better placed to survive the weaning process which is a critical first step to commercially culturing glass eels. In our experiment glass eels that failed to wean were observed to gradually decline in condition, becoming thinner in comparison to those feeding. Similar descriptions of decline in eel condition have been documented in the shortfin eel (Jellyman 1979; Chisnall *et al.* 2002) and the European eel (Rodriguez *et al.* 2005) with cause of death being similarly being attributed to starvation.



For fishery managers, better knowledge of first year glass eel mortality and subsequent recruitment may improve the management of eel fisheries and assist our understanding of this critical life stage (Jessop 1998). The prediction of year class strength, therefore, would be aided by research that focuses on the condition of glass eels around the time of freshwater migration but prior to recruitment into the adult population. For example, it may be practical or desirable to more highly exploit a portion of the glass migration where natural mortality may be higher. Overall exploitation by a glass eel fishery might, amongst other considerations, be set relative to the first-year glass eel mortality rate.

In conclusion, our results suggest there is a link between glass eel size (length and dry mass), quality (lipid content) and subsequent growth and survival in captivity. It would therefore be prudent for aquaculturists to harvest early season glass eels and/ or sort glass eels by length and retain the larger individuals which will return higher survival and growth rates in culture.

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